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Multilocus, DNA-based phylogenetic analyses reveal three new species lineages in the *Phellinus gabonensis–P. caribaeo-quercicola* species complex, including *P. amazonicus* sp. nov.

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Abstract: Species complexes in the poroid Hymenochaetaceae are well documented in the temperate areas. Potential species complexes are less known in tropical areas, however. In the last ten years, four phylogenetically and morphologically closely related species of *Phellinus* (Hymenochaetaceae) were described from various tropical/subtropical areas viz. *P. caribaeo-quercicola*, *P. gabonensis*, *P. ellipsoideus*, and *P. castanopsidis*. They are characterized by cushion-shaped basidiomata, ventricose, commonly hamate hymenial setae, and broadly ellipsoid, thick-walled, pale yellowish basidiospores. Pursuing the studies of this complex, a phylogenetic approach based on DNA sequence data from the nuc rDNA regions ITS1-5.8S-ITS2 (ITS) and partial 28S (including the domains D1, D2, D3) and on part of the translation elongation factor 1-α (*tef1*, region between exons 4 and 8) revealed three new lineages or phylogenetic species. Two of these phylogenetic species are composed of exclusively on Neotropical specimens. One of them, described below as *Phellinus amazonicus* sp. nov., is represented by multiple collections originating from Neotropical, lowland, dense, moist forest at the western edge of the Amazon Basin in Ecuador, the Guiana Shield in French Guiana and (more likely) Trinidad. The second Neotropical phylogenetic species is represented in our phylogenetic analyses by a single collection from northeastern Argentina. It is also potentially known from two herbarium specimens originating from southern Brazil, for which no sequence data is available. It is left for now as *Phellinus* sp. 1, waiting to gather more specimens and DNA sequences data. The third new phylogenetic species is known by a single collection (pure culture) of uncertain origin. It is thought to represent *Phellinus setulosus*, a Southeast Asian taxa. From an evolutionary perspective, tree species occurring in the Neotropics (*P. amazonicus*, *P. caribaeo-quercicola*, and *Phellinus* sp. 1) have a closely related genetic background and form a well supported Neotropical lineage.

Key words: biogeography, Hymenochaetaceae, *Phellinus*, polypores, North America

Introduction

Phellinus[3] is one of the major genera of Hymenochaetaceae (Basidiomycota) (Ryvarden and Johansen 1980, Larsen and Cobb-Poulle 1990, Corner 1991). In the last three decades, it was repeatedly shown to be polyphyletic (Wagner and Fischer 2002, Larsson et al. 2006) and consequentially, many species were redistributed into multiple morphologically more homogeneous and phylogenetically monophyletic entities, which were worth recognition at the generic level.

Phellinus, related genera and more globally the poroid Hymenochaetaceae are also well known for containing (morphological) species complexes. These complexes encompass a number of taxonomic entities for which divergences (genetic, biological, and ecological) occurred independently of tangible morphological changes (e.g. Fischer and Binder 2004, Tomšovský et al. 2010a, b). Therefore, delimiting these entities using morphological features proved challenging. Additional descriptors were tentatively considered to circumscribe taxa, including, for example, autecological requirements such as the host relationships (preference/specificity). However, the pertinence of these descriptors for characterizing taxonomic entities in the Hymenochaetaceae also has been variously debated (e.g. Pieri and Rivoire 2000, Rizzo et al. 2003, Tomšovský et al. 2010a, b). Their validation is almost case-by-case.

Application of the phylogenetic species concept, using the principle of multiple gene genealogy concordance

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[3] Authorships of scientific names are reported (Table I).

(Taylor et al. 2000, 2006), helped to evidence diversity within complexes of Hymenochaetaceae (e.g. Decock et al. 2007; Amalfi et al. 2010, 2012, 2014; Amalfi and Decock 2013, 2014; Vlasák and Kout 2011; Zhou et al. 2015). A posteriori, it may validate the pertinence of ecological descriptors and help to circumscribe the (bio)geographical distribution range of the various taxa (Amalfi et al. 2012). As suggested by Amalfi et al. (2012), integrating morphological, ecological, biogeographical, and DNA sequence data could yield a more complete (holistic) concept of the species; this is the objective of the "consolidated species concept" recently highlighted within Ascomycota (Quaedvlieg et al. 2014).

For historical reasons, species complexes within the Hymenochaetaceae are mainly documented in northern temperate areas (e.g. Niemelä 1975; Tomšovský et al. 2010a, b; Vlasák and Kout 2011; Amalfi et al. 2012). Species complexes are much less documented in the tropical areas, which are still critically underexplored (Yombiyeni et al. 2011). Nonetheless, concerning the Neotropics, Decock et al. (2007); Amalfi and Decock (2013, 2014) and Campos-Santana et al. (2014) showed the existence of multiple phylogenetic species within the presumed *Fomitiporia punctata* (P. Karst.) Murrill or *Fomitiporia robusta* (P. Karst.) Fiasson & Niemelä complexes. Decock et al. (2013) and Yombiyeni et al. (2015) also showed complex pictures within the *Phylloporia spathulata* (Hook.) Ryvarden and the *Phylloporia pectinata* (Klotzsch) Ryvarden species concept with multiple lineages, indicating multiple species. Tian et al. (2013); Vlasák et al. (2013) and Zhou et al. (2015) also demonstrated that *I. linteus* (Berk. & M.A. Curtis) Teixeira sensu auctores (e.g. sensu Ryvarden 2004, under *Phellinus linteus* [Berk. & Curt.] Teng) encompassed several cryptic species in the Neotropics, recognized by molecular data.

In the last decade, four phylogenetically closely related species of *Phellinus* were described from various tropical/subtropical areas. They are, chronologically, *P. caribaeo-quercicola* (Decock et al. 2005), *P. gabonensis* (Yombiyeni et al. 2011), *P. ellipsoideus* (Cui and Decock 2013) (originally described as *Fomitiporia ellipsoidea* B.K. Cui & Y.C. Dai, Cui and Dai 2008) and *P. castanopsidis* (Cui and Decock 2013). These species are morphologically very similar, sharing cushion-shaped basidiomata, ventricose, apically curved to hamate hymenial setae, and broadly ellipsoid, slightly thick-walled basidiospores; hence, they form a morphological species complex. However, they have either different ecological requirements, in terms of hosts and habitats, and/or disjoint distribution ranges. *Phellinus caribaeo-quercicola* originates from the Caribbean and is known so far exclusively on *Quercus* L. (Fagaceae) (Decock et al. 2005; Vlasák et al. 2011); *Phellinus gabonensis* is known from various angiosperms in the western edge of the Guineo-Congolian Forest, in Gabon (Yombiyeni et al. 2011); *Phellinus ellipsoideus* and *P. castanopsidis* are known from southern tropical/subtropical China, the former on unidentified angiosperms and the latter on *Castanopsis* (D. Don) Spach (Fagaceae) (Cui and Dai 2008; Cui and Decock 2013). In a phylogenetic perspective, these four species have a monophyletic origin and form a distinct lineage within *Phellinus* (Decock et al. 2007; Yombiyeni et al. 2011; Cui and Decock 2013). Decock et al. (2005). Yombiyeni et al. (2011) suggested that these species have affinities with *Phellinus setulosus* sensu Corner (1991).

Pursuing the studies of this complex, the taxonomic status of a set of specimens from Argentina, Ecuador, French Guiana, and southern Brazil was questioned. The specimens from Brazil were first identified to *P. gabonensis* based on gross morphological similitude, at macro- and microscopic levels (Campos-Santana and Borges da Silveira 2011). Nonetheless, phylogenetic inferences based on DNA loci (the nuc rDNA regions ITS1-5.8S-ITS2 [ITS] and partial 28S (including D1–D3 domains) and part of the translation elongation factor 1-α (*tef1*, region between exons 4 and 8]), showed that these collections are distributed over two distinct lineages of which none is equated to *P. gabonensis*. They are therefore interpreted as belonging to two distinct phylogenetic species. One of them is described below as *Phellinus amazonicus* sp. nov., whereas the second is left for now as *Phellinus* sp. 1.

The phylogenetic inferences have also shown that a living culture received from CFMR as *Fuscoporia wahlbergii* is related to this complex, forming a separate lineage. This lineage is discussed in relation to *P. setulosus*.

MATERIALS AND METHODS

Collection localities.—MUCL materials of the new taxon were collected in several Neotropical lowland, dense, moist forest (broadleaf evergreen rainforest) ecotypes (Eva et al. 1999).

The majority of the specimens originated from the wet (humid to very humid) evergreen lowland forest of the Guiana Shield in French Guiana (FG) (Granville 1988) and from the western edge of the Amazon Basin in Ecuador. In French Guiana, specimens originated from the Nouragues Natural Reserve, CNRS Inselberg and Pararé research plots (approx 04°05N, 52°41′W and 04°02′N, 52°41′W, respectively) and the hills of the Maroni River valley, western FG (approx 02°732′N, 54°012′W, elevation approx 120–240 masl). The annual average of precipitation at the CNRS research plots is approx of 3000 mm. In FG, specimens were also collected in drier littoral forests (approx 05.215N, 052.909W) of the *Dimorphandra* type, developed on detrital sandy soils and commonly designated as "white sand forests" (de Granville 1986, 2002), in which *Dimorphandra polyandra*

Benoist (Caesalpiniaceae) is the dominant large-trunked tree species.

In Ecuador, specimens originated from the Yasuni Biosphere Reserve in the neighboring Yasuni Biological Station, including the Center for Tropical Forest Science–Global Earth Observatories (CTFS-ForestGEO) forest monitoring plot (approx 0°40.17S, 76.0W). Locally, the annual average precipitation is approx 3080 mm (Valencia et al. 2004).

Specimens from Brazil were collected in the Atlantic Forest (rainforest phytogeographical area) in Rio Grande do Sul, Dom Pedro de Alcântara, RPPN do Professor Luis Baptista (approx 29.37S, 49.84W) and Santa Catarina, Itapuá, RPPN Volta Velha (approx 26.68S, 48.25W). The Atlantic rainforest is characterized by a warm and humid climate, with rainfall distributed throughout the year and a hot summer (Peel et al. 2007).

Specimens.—Herbarium specimens are preserved at MUCL with a duplicate at CAY (specimens from French Guiana, isotype), PUCE (specimens from Ecuador), CORD (specimen from Argentina), ICN (specimens from Brazil) and NY (type of the new species, specimens from Trinidad) (herbarium acronyms are from Thiers [continuously updated]).

MUCL strains were isolated from basidiomata tissues during fieldwork, using malt extract agar with 2 ppm benomyl (benlate) and 50 ppm chloramphenicol, and later when necessary, purified from bacteria in the laboratory. Living cultures (strains) are preserved at MUCL and in the culture collection of ECOFOG, Kourou. The ex-holotype strain is also deposited at CBS. The strain CFMR-CSIRO-5816 (MUCL 54670) was received from CFMR (USDA, Madison, Wisconsin), courtesy of Dr K. Nakasone.

Morphology and anatomy.—Basidiomata colors are described according to Kornerup and Wanscher (1981). Specimens were examined in Melzer's reagent, Lactophenol Cotton Blue (Kirk et al. 2001) and KOH 4%. All microscopic measurements were done in Melzer's reagent. In presenting the size range of the microscopic elements, 5% of the measurements was excluded from each end and are given in parentheses when relevant. Ave = arithmetic mean, Q = length-to-width ratio of basidiospores, and ave_Q = arithmetic mean of the ratio Q. Thirty elements per specimen were measured for each microscopic character.

Sequencing.—DNA extraction, amplification, and sequencing of the 5′ end of the nuclear ribosomal 28S (including domains D1, D2, and D3, hereafter 28S) and ITS regions (including ITS1, ITS2, 5.8S), and partial *tef1* gene (region located between exons 4 and 8) were described in Yombiyeni et al. (2011) and Amalfi and Decock (2013). Sequencing reactions were performed at Macrogen Ltd. (Seoul, Korea) with the primers LROR, LR3, LR3R, LR5 for the 28S; ITS1, ITS2, ITS3, ITS4 for the ITS (http://biology.duke.edu/fungi/mycolab/primers.htm, White et al. 1990); and 2212R, 1953R, 983F, 2218R for the *tef1* (Rehner and Buckley 2005, Matheny et al. 2007).

Phylogenetic analyses.—The dataset used in the present study to infer phylogenetic inferences is the same used previously by Yombiyeni et al. (2011), implemented with collections from South America and Asia (TABLE I). The methodologies and parameters for running phylogenetic analyses were described in detail in Yombiyeni et al. (2011) and are summarized below. Alignment is deposited at TreeBASE (http://www.treebase.org/treebase/index.html) under S19196. Forty-one collections representing eight species/potential species were included in the phylogenetic analysis (TABLE I). Nucleotide sequences were automatically aligned with ClustalX 2.0.11 (Thompson et al. 1997) then manually adjusted by direct examination with Se-Al 2.0a11 (http://tree.bio.ed.ac.uk/software/seal). The assignment of codon positions was confirmed by translating nucleotide sequences into predicted amino acid sequences with MacClade 4.0 (Maddison and Maddison 2000). Potential ambiguously aligned segments were detected by gBlocks 0.91b (Castresana 2000; http://molevol.cmima.csic.es/castresana/Gblocks.html, settings "allow smaller final blocks", "allow gaps within blocks"). *Phellinus* sp. MUCL 52000 was designated as outgroup (Yombiyeni et al. 2011).

Phylogenetic analyses were performed separately for each gene region using maximum parsimony (MP) as implemented in PAUP* 4.0b10 (Swofford 2003), Bayesian inference (BI) as implemented in MrBayes 3.2.1 (Ronquist et al. 2012) and Maximum likelihood (ML) as implemented in RAxML 7.0.4 (Stamatakis 2008).

For MP analyses, gaps were treated as missing. The most parsimonious trees (MPT) for each dataset were identified using heuristic searches with 1000 random addition sequences, further evaluated by bootstrap analysis, retaining clades compatible with the 50% majority-rule in the bootstrap consensus tree. Analysis conditions were: tree bisection addition branch swapping (TBR), starting tree obtained via stepwise addition, steepest descent not in effect, MULTREES effective. A bootstrap support value (BS) above 85% was considered supported by the data.

Evolution models for Bayesian inference were estimated using the Akaike information criterion (AIC) as implemented in jModeltest 3.7 (Posada and Crandall 1998). The best-fit models for each partition were implemented as partition-specific models within partitioned mixed-model analyses of the combined dataset. In order to facilitate the data partitioning by codon position for the *tef1*, the three introns present were excised and analyzed as a distinct partition.

Bayesian analyses were implemented in two independent runs, each with four simultaneous independent chains for 10 000 000 generations for each dataset, starting from random trees, and keeping one tree every 1000th generation. All trees sampled after convergence (ave. standard deviation of split frequencies <0.01 and confirmed using Tracer 1.4 (Rambaut and Drummond 2007) were used to reconstruct a 50% majority-rule consensus tree (BC) and to estimate posterior probabilities. The Bayesian posterior probability (BPP) of each node was estimated based on the frequency at which the node was resolved among the sampled trees with the consensus option of 50% majority-rule (Simmons et al. 2004). A BPP above 0.95 was considered a significant value.

Maximum likelihood (ML) searches conducted with RAxML involved 1000 replicates under the GTRGAMMAI

TABLE I. List of species, collections and sequences used in the phylogenetic analyses

Genus/species	GenBank accession No.		
Voucher specimens/cultures reference	28S	ITS	*tef1*
Phellinus amazonicus Campos-Santana & Decock			
MUCL 51476	KU376295	KU499930	KU936768
MUCL 51478	KU376294	KU499929	KU936769
MUCL 51483	KU376308	KU499943	KU936770
MUCL 51487	KU376307	KU499942	KU936771
MUCL 53036 (T)	KU376305	KU499940	KU936772
MUCL 53056	KU376302	KU499937	KU936773
MUCL 53084	KU376301	KU499936	KU936774
MUCL 53095	KU376296	KU499931	KU936775
MUCL 53117	KU376304	KU499939	KU936776
MUCL 53128	KU376303	KU499938	KU936777
MUCL 53141	KU376300	KU499935	KU936778
MUCL 53685	KU376309	KU499944	KU936779
MUCL 55050	KU376297	KU499932	KU936780
MUCL 55075	KU376298	KU499933	KU936781
MUCL 55076	KU376299	KU499934	KU936782
Phellinus caribaeo-quercicola Decock & S. Herrera			
MUCL 46003	DQ127279	HM635697	HM635725
MUCL 46004 (T)	DQ127280	HM635698	HM635726
MUCL 46005	DQ127281	HM635699	HM635727
Phellinus castanopsidis Cui & Decock			
CUI 10153	JQ837944	JQ837956	KU936783
CUI 10157	JQ837945	JQ837957	KU936784
Phellinus ellipsoideus (B.K.Cui & Y.C.Dai) B.K.Cui, Y.C.Dai & Decock			
MUCL 45929	DQ127283	KU954544	KU936785
MUCL 47867	KU954540	KU954545	KU936786
MUCL 47820	KU954542	JQ837952	KU936787
MUCL 47822	KU954543	JQ837954	KU9367788
MUCL 47866	KU954541	JQ837953	KU9367789
Phellinus gabonensis Decock & Yombiyeni			
MUCL 47562	HM635682	HM635721	HM635734
MUCL 51275	HM635683	HM635720	HM635735
MUCL 51277	HM635684	HM635719	HM635736
MUCL 52007	HM635685	HM635718	HM635729
MUCL 52012	HM635687	HM635717	HM635730
MUCL 52014	HM635688	HM635716	HM635728
MUCL 52023 Clone A	HM635689	HM635700	HM635732
MUCL 52023 Clone H	HM635689	HM635707	HM635732
MUCL 52025 Clone A (T)	HM635690	HM635708	HM635731
MUCL 52025 Clone H	HM635690	HM635715	HM635731
MUCL 52070	HM635686	HM635722	HM635733
Phellinus sp. 1			
MUCL 51334	KU954538	KU954539	KU936790
Phellinus cf. *setulosus* (Lloyd) Imazeki			
MUCL 54670[1]	KU954536	KU954537	KU936791

TABLE I. Continued

Genus/species	GenBank accession No.		
Voucher specimens/cultures reference	28S	ITS	*tef1*
Phellinus spiculosus (W.A. Campb. & R.W. Davidson) Niemelä			
MUCL 53634	KU376306	KU499941	KU936792
Phellinus sp.			
MUCL 52000	HM635695	HM635723	HM635737
MUCL 52001	HM635696	HM635724	HM635738

T, PT = type, paratype.
[1] Courtesy of Dr. K. Nakasone, CFMR, USA.

model, with all model parameters estimated by the program. In addition 1000 bootstrap (ML BS) replicates were run with the same GTRGAMMAI model. We provided an additional alignment partition file to force RAxML software to search for a separate evolution model for each dataset. Clades with Maximum likelihood bootstrap values of 85% or greater were considered supported by the data.

RESULTS

Phylogenetic analyses.—The 5′ region of the 28S gene including domains D1, D2, and D3 ranged from 1039 (*Phellinus* sp., MUCL 52000, MUCL 52001) to 1056 bps (*P. castanopsidis*, CUI 10153, CUI 10157). The ITS1-5.8S-ITS2 region ranged from 537 (*P. castanospidis* CUI 10153, CUI 10157) to 602 bps (*P. gabonensis* MUCL 52023).

Amplicons ranging from 1156 (*P. setulosus* MUCL 54670) to 1162 bps (*Phellinus* sp. MUCL 52000, MUCL 52001) were obtained from the partial *tef1*. This region contains three clade specific, confidently alignable introns.

The final DNA sequence alignments resulted in 1079 characters, including gaps, for the 28S gene (47 variable parsimony-uninformative and 58 parsimony-informative positions), 700 for the ITS dataset (49 variable parsimony-uninformative and 165 parsimony-informative positions), 943 for the *tef1* exons partition (22 variable parsimony-uninformative and 177 parsimony-informative positions), and 283 for the *tef1* introns dataset (23 variable parsimony-uninformative and 107 parsimony-informative positions).

By comparing MP bootstrap tree topologies obtained for the individual datasets, no conflict involving significantly supported nodes was found; the datasets were therefore combined.

Combined dataset analysis.—3008 characters were included in the analysis of which 141 were variable but parsimony-uninformative and 507 parsimony-informative.

The heuristic search produced 9570 equally most parsimonious trees (928 steps long; CI = 0.825, RI = 0.920, RC = 0.760), distributed between two main topologies. These topologies differ only by the position of CFMR-CSIRO-5816 strain.

The best-fit models selected were: GTR+G for the ITS-5.8S dataset with unequal base frequencies (A = 0.2264, C = 0.2009, G = 0.2567, T = 0.3161) and a gamma distribution shape parameter (γ) of 0.3107; GTR+I+G to the 28S dataset (A = 0.2538, C = 0.2171, G = 0.2965, T = 0.2326) a proportion of invariable sites (I) of 0.7001 and a γ of 0.6936; GTR+G for the *tef1* exons first codon position (A = 0.2430, C = 0.2091, G = 0.3316, T = 0.2163, γ = 0.2475); F81+I for the *tef1* exons second codon position (A = 0.3119, C = 0.2506, G = 0.2351, T = 0.2024, I = 0.9658); HKY+G for the *tef1* exons third codon position (A = 0.1631, C = 0.3123, G = 0.2203, T = 0.3043, γ = 0.6288); HKY+G for the *tef1* introns dataset (A = 0.2569, C = 0.2128, G = 0.2066, T = 0.3237, γ = 0.6938).

The two Bayesian runs converged to stable likelihood values after 1920 generations and 8080 (about 80% of the sampled trees) stationary trees from each analysis were used to compute a 50% majority rule consensus tree in PAUP* to calculate posterior probabilities.

In the ML searches with RAxML the combined dataset alignment had 772 distinct patterns with a proportion of gaps and undetermined characters of 9.48%.

The BC tree (FIG. 1) and the best scoring ML tree (tree score of −lnL = −8436.061016) were mostly identical to the strict consensus of the 9570 equally most parsimonious trees. The topology of this tree is highly concordant with topologies obtained analyzing the individual datasets.

As shown (FIG. 1), the phylogenetic inferences resolved the collections from French Guiana and Ecuador as a distinct, well supported lineage. This lineage is closely related and sister to the *P. caribaeo-quercicola* clade and distant from the *P. gabonensis* clade.

The collection from Argentina (*Phellinus* sp. 1, MUCL 51334) is also resolved as a distinct lineage, subtending the *P. caribaeo-quercicola*–(FG-EC) clade.

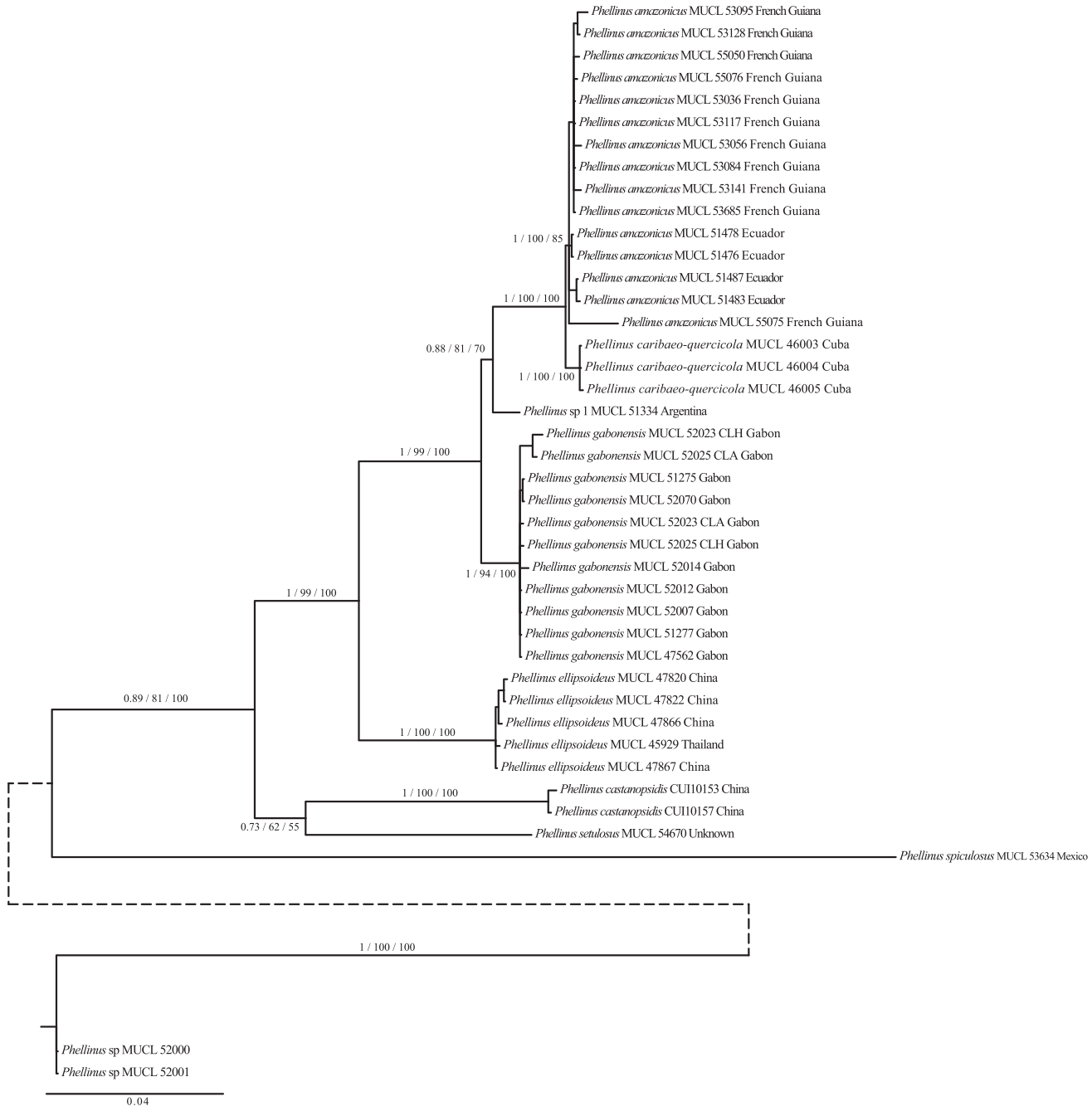
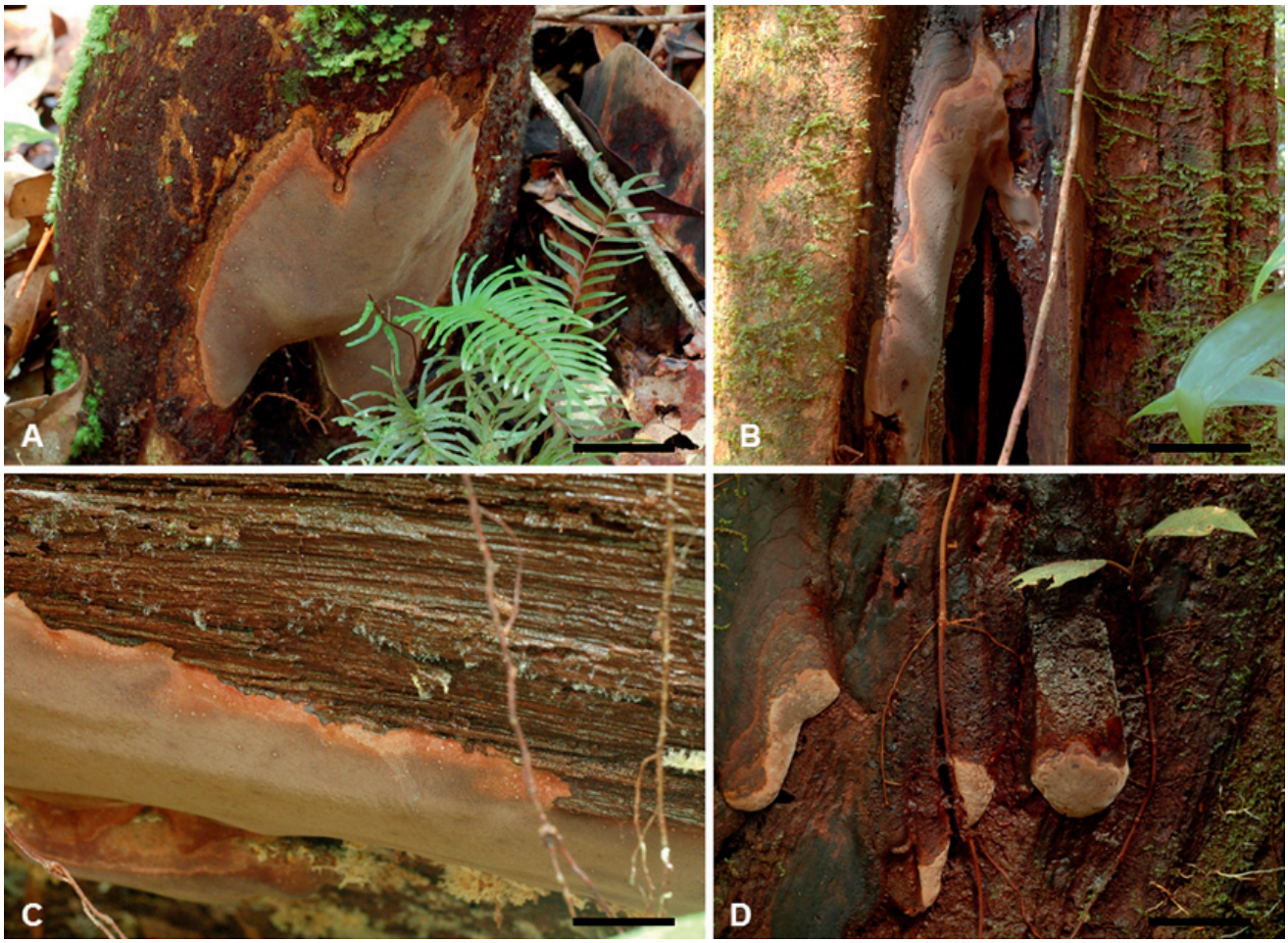


FIG. 1. The 50% majority-rule consensus tree from Bayesian inference of the combined nuclear ITS, 28S and *tef1* sequences. Thickened branches in bold indicate bootstrap support greater than 70% and Bayesian posterior probability greater than 0.95. For selected nodes, parsimony bootstrap support value and Bayesian posterior probabilities are, respectively, indicated to the left and right of slashes.

The single strain CFMR-CSIRO-5816 (MUCL 54670) forms a third new lineage, which is either sister to *P. castanopsidis* clade or basal and isolated.

Morphological analysis.—Morphologically, the collections examined from Ecuador (6) and French Guiana (21) are very homogeneous as far as their basidiomata, hyphal system, vegetative hyphae differentiation, hymenial setae shape and size and basidiospores are concerned. Two collections from Trinidad share these characters. The vegetative hyphae are short skeletal hyphae of limited growth as described in *P. gabonensis* (Yombiyeni et al. 2011) and also in *P. setulosus* (Corner 1991). The collections from French Guiana and Ecuador could not be differentiated from each other based on macro- or microscopic characters.



FIGS. 2A–D. *Phellinus amazonicus* basidiomata in situ. A. Young basidiomata (MUCL 55075). B. Mature basidiomata on a vertical substrate (MUCL 53084). C. (MUCL FG-09-125). D. Mature, pending, obclavate basidiomata (MUCL53056). Scale bars: A, B = 10 cm; C, D = 5 cm.

The unique collection from Argentina (MUCL 51334) shares most of these morphological features described above. It differs in developing larger and continuous pulvinate sheets on dead trees. Two additional specimens from Brazil, for which no sequence are presently available, are very similar.

Taxonomic conclusions.—The specimens from French Guiana and Ecuador (and possibly Trinidad) are interpreted as an undescribed species, which is described below as *Phellinus amazonicus*. The collection from Argentina is also interpreted as a distinct species. Two additional collections from Southern Brazil might represent this species, but for now no sequence could be obtained from their voucher herbarium specimens. Therefore, due to the paucity of material currently available, we refrain to describe it.

Although with conspicuous basidiomata (FIGS. 2, 3) and, in the case of *P. amazonicus*, locally common, no name could be found for these two species within *Phellinus* (Larsen and Cobb-Poulle 1990, Ryvarden 2004).

Nonetheless, in the frame of our revision of Neotropical Hymenochaetaceae (Decock et al. 2006, 2007, 2013; Amalfi and Decock 2013, 2014; Campos Santana et al. 2014, 2015), *Pyropolyporus robinsoniae* Murrill (Murrill 1907) came to our attention. *Pyropolyporus robinsoniae* is currently accepted as a synonym of *Fuscoporia wahlbergii* (Fr.) T. Wagner & M. Fisch. (Larsen and Cobb-Poulle 1990, Ryvarden 1990). The revision of its type specimen (JAMAICA. Monkey Hill, decaying root tree, 11 Jul 1904, *W. J. Robinson s.n.*, NYBG 00743008) confirmed its main features, viz. pileate basidiomata with concentrically sulcate, shortly tomentose pileus and, microscopically, long and narrow (subulate), apically hamate to hooked setae. All these characteristics indeed point toward *F. wahlbergii* (Ryvarden 2004, see below). *Pyropolyporus robinsoniae* could thus not apply to our species from French Guiana and Ecuador.

FIGS. 3A–D. *Phellinus amazonicus* basidiomata in situ. A. General view of the microhabitat (arrows indicate basidiomata) (MUCL55326). B. Mature basidiomata (MUCL 55283). C. Mature, massive basidiomata extending into a hollowed trunk (MUCL53117). D. Details of the pocket, alveolar rot (MUCL55627). Scale bars: A = 30 cm; B, C, D = 10 cm.

Nevertheless, during the study of Neotropical specimens of *F. wahlbergii/P. robinsoniae*, we came across two collections originating from Trinidad that were once identified to *P. robinsoniae*. The specimen labeled 3065 (NY, cf. list of specimens below) was anonymously annotated "abnormally developed (*Pyropolyporus robinsoniae*)" and annotated by Lowe (1957, date on label) as "*P. robinsoniae*". Both specimens show the same combination of macro- (thickly pulvinate basidiomata) and microscopic characters (short skeletal hyphae, ventricose, curved hymenial setae, yellowish, broadly ellipsoid basidiospores) that observed in our specimens of *P. amazonicus* from French Guiana and Ecuador. These two specimens, subject to confirmation by sequencing, more likely they belong to the latter species.

The third new lineage is composed by a single entry viz. the culture CFMR/CSIRO-5816, (USDA, Madison, Wisconsin) received under the name of *F. wahlbergii*. This culture is of uncertain origin, however; it was very likely received at CFMR from CSIRO (Commonwealth Scientific and Industrial Research Organization, Australia), but there is no data about the locality of the original voucher specimen.

CFMR/CSIRO-5816 (cf. above) was also examined in the frame of a revision of *F. wahlbergii* and of its synonyms. *Fuscoporia wahlbergii* is, among other features (see above), characterized by pileate basidiomata and subulate, straight to commonly apically hamate to hooked hymenial setae. The identification of CFMR/CSIRO-5816 to *F. wahlbergii* could thus indicate a voucher specimen with hamate to hooked setae and perhaps a pileate basidiomata. However, given that phylogenetic inferences showed this culture to be affine to *P. ellipsoideus* and *P. castanopsidis* (FIG. 1) (but forming a distinct lineage, indicative of a distinct species) and not to *F. wahlbergii* (which belongs to the distant *Fuscoporia* lineage, Larsson et al. 2006), we suggest it might belong to *P. setulosus*; this species also is characterized by hamate to hooked setae and a pileate basidiomata (Cunningham 1965, Corner 1991). Nevertheless, this hypothesis should be ascertained by examination of the voucher specimen or gathering other DNA

sequence data from other *P. setulosus* specimens, which identification is confirmed.

More globally, this case (*F. wahlbergii* vs. *P. cf. setulosus*) and the preceding (*F. wahlbergii/P. robinsoniae* versus *P. amazonicus*) indicate confusions between species of the *P. caribaeo-quercicola* complex and *F. wahlbergii*.

TAXONOMY

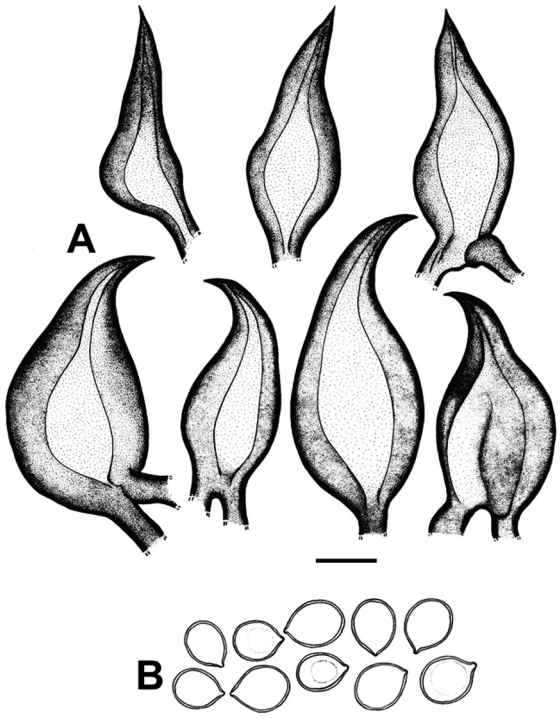
Phellinus amazonicus Campos-Santana & Decock sp. nov. FIGS. 2–5

MycoBank MB816308

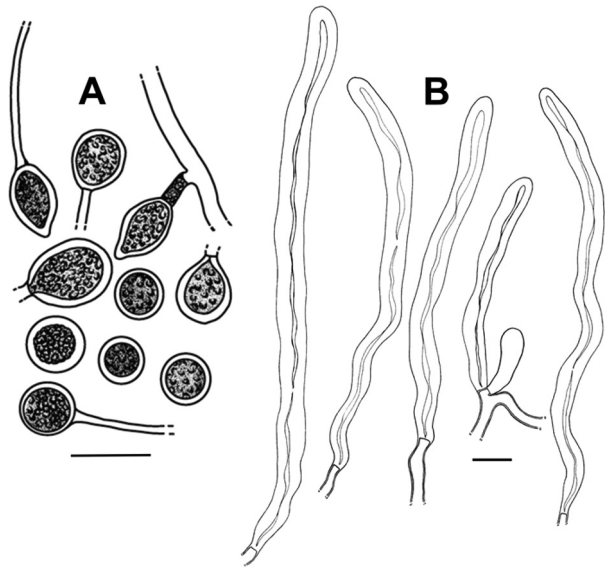
Typification: FRENCH GUIANA. Municipality of Regina, Nouragues Natural Reserve, CNRS "inselberg" Research Station, approx 4°05′N–52°41′W, Grand Plateau, dead fallen hollowed, trunk, basidiomata covering the internal wall, 28 Jul 2010, *C. Decock, FG-10-136* (**holotype** NY 02674262, **isotype** MUCL 53036). Extype culture MUCL 53036.

Etymology: "*amazonicus*" (L.), from the Amazon rainforest.

Diagnosis: Phellinus amazonicus is characterized by cushion-shaped basidiomata, a dimitic hyphal system with short skeletal hyphae, ventricose hymenial setae, the apex curved to hamate, broadly ellipsoid basidiospores averaging 5.0 × 4.3 µm and occurring in the Neotropical rainforest (of eastern Ecuador and French Guiana). Basidiomata perennial, resupinate to effused when young, following the surface of the substrate, soon cushion-shaped to thickly cushion-shaped, up to nodulose to obclavate when growing on vertical substrates, strongly adnate and very difficult to cut off from the substrate; individual cushion extending up to 600 mm in the longest dimension (cushions growing side by side may also fuse, resulting in large compound basidiomata extending over longer surface, >1 m), 50–200 mm wide, from 3 mm thick in young specimens up to 100 mm thick in the thickest part of multilayered specimens; consistency very hard, woody; margin 0.5–4 mm wide, densely and very minutely velutinous when young, (whitish) to yellowish brown (5E[7–8]) at the margin, turning light brown to rusty (6E[7–8]), the marginal areas progressively indurating in old, pulvinate, multilayered specimens, and gradually mimetic with the surrounding wood; pore surface light to dark brown (6F[4–7]), chocolate brown (6E[4–8], leather, cocoa brown), glancing with light, then appearing paler, light to golden brown (6D[3–4], camel to cafè au lait); pores regular, round to ellipsoid when growing on standing trunk, 7–10/mm, (75–) 90–130 (–140) µm diam (ave = 102 µm); dissepiments entire, thin to thick, (20–) 25–75 µm (ave = 39 µm); subiculum very thin to almost absent, negligible compared to the thickness of the tube layers, concolorous with the older tube layer; tube layer single to multiple, with numerous individual, weakly distinct layers in old specimens, each 2–35 mm thick, totaling up to 100 mm thick, brown to dark brown (6[E–F]6–7, cocoa brown to burnt amber), the older layers light brown (6D[5–6], sunburn to camel, 7F[4–5]). Hyphal system dimitic (of the fifth degree, Corner 1991), identical in the context and hymenophoral trama; generative hyphae



FIGS. 4A–C. *Phellinus amazonicus* (MUCL 51487). A. Hymenial setae. B. Basidiospores. Scale bars: A, B = 5 µm.



FIGS. 5A–B. *Phellinus amazonicus*, MUCL 51487. A. Chlamydospores in in-vitro culture. B. Skeletal hyphae, basidiomata. Scale bars: A = 14 µm, B = 5 µm.

hyaline to yellowish, thin-walled, slightly branched, 1.5–3 µm wide; vegetative hyphae as skeletal hyphae of limited growth, (30–) 47–100 (–125) µm long (ave = 57 µm), 2.0–2.5 µm diam at the basal septum to 2.5–3.0 (–4 µm) diam (ave = 2.6 µm) in the main part, densely packed, with subparallel orientation, straight or occasionally geniculated, especially near the base, then with small, lateral aborted process pale brown, ending rounded, thick-walled but with the lumen open.

Hymenium: basidia barrel-shaped, 6.5–8.5 × 4.0–6.0 µm, with four small sterigmata; basidioles subglobose to barrel-shaped; basidiospores (ellipsoid to) broadly ellipsoid, thin-walled and hyaline first, distinctly thick-walled, faintly yellowish when mature, pale creamy in dense basidiospores print, 0–1 guttate, negative in Melzer's reagent, 4.5–5.5 (–6.0) × (3.5–) 4.0–5.0 (–5.5) µm (ave = 5.0 × 4.3 µm) R = 1.0– 1.45 (ave_R = 1.2); hymenial setae always present, scattered to commonly abundant, mono-, bi-, or occasionally three-rooted, occasionally with a small hyphal-like base, acuminate to symmetrically or unilaterally ventricose, straight to curved, occasionally slightly sinuous, the apex acute, straight to commonly curved, or slightly (rarely strongly) hamate, hooked, (13.0–) 15.0 – 23.0 (–25.0) × (4.5–) 5.0–9.5 (–10.0) µm (ave = 17.2 × 6.6 µm); chlamydospores absent in basidiomata (but produced in in vitro culture, variably abundant subglobose to globose, thick-walled, hyaline to brownish, 8.0–15.0 × 8.0–11.0 µm (ave = 10.9 × 9.5 µm), R = 1.0– 1.50 (ave_R = 1.15).

Type of rot: white pocket (alveolar) rot (FIG. 3H).

Ecology (substrate, host, habitat): known from dead, fallen or more rarely standing trunks, or stumps, once found growing at the base of a living tree, usually large (commonly ≥ 50 cm diam), underneath, on the side, or covering internal walls of hollowed trunks or stumps, on various angiosperms of which *Dimorphandra polyandra* (Caesalpiniaceae) and *Minguartia guianensis* Aublet (Olacaceae), humid to very humid rainforest of the Guiana Shield and of the western Amazonia, and in seasonally drier, coastal forest on detrital white sands in French Guiana.

Distribution: Known from the Northeastern Guiana Shield, French Guiana, western edge of the Amazonia, Ecuador and (subject to confirmation by sequence data) in the insular Trinidad.

Specimens examined. (SUPPLEMENTARY DATA 1).

Notes.—Phellinus amazonicus is well characterized by commonly thickly, well delimited, cushion-shaped basidiomata, extending horizontally, or downward when growing on a vertical substrate, and which indurated margins are hardly distinguished from the surrounding wood. Additionally, it is characterized by a dimitic hyphal system with short skeletal hyphae, ventricose, apically curved to hamate hymenial setae, and broadly ellipsoid, pale yellowish, slightly thick-walled basidiospores.

Ecologically, the species grows mostly on dead, standing or fallen trunk, or stumps. It was once found growing on a living tree. It has (very likely) no direct host specificity or preference but a substrate specificity. The known host range includes *Dimorphandra polyandra* and *Minguartia guianensis*, but it is incompletely known; the nature of the most common substrate (dead fallen trunks) makes its botanical identification (in situ) difficult in most cases. Nonetheless, locally, the species may have an indirect host preference linked to the substrate specificity. The biological characteristics of the species, of which the reproduction strategy, involve large, perennial and dense basidiomata. The development of such basidiomata should require large amount of resources that could only be obtained through colonization of large substrates; hence, the basidiomata are found mostly on large tree of a diam ≥50 cm. This substrate specificity might induce indirect host specificity; hence, in the coastal white sand forest in FG, *P. amazonicus* was found mostly on the legume *D. polyandra*, which is locally also the dominating large-trunked species.

The species could be locally common. It has been repeatedly observed in the research plots of the Nouragues Research Station in FG, both within the approximate 120 Ha of the "Inselberg" grand and petit plateau and "Pararé" camp. (SUPPLEMENTARY DATA 1). It has been also repeatedly observed in the 50 Ha parcels of the CTFS-ForestGEO Yasuni monitoring plot, Ecuador (SUPPLEMENTARY DATA 1).

In a biogeographical perspective, *P. amazonicus* is currently known from the wet (humid to very humid) evergreen lowland forest of the Guiana Shield and of the eastern Amazonia. It also occurs very likely in the insular Trinidad, north of Venezuela. Its distribution between these two locations and its northern and southern limits are unknown. No distribution pattern could be defined for now; the species might have a pan-Amazonian or alternatively, a northern, peri-Amazonian distribution, which would be worth searching for.

In both phylogenetic and biogeographical perspectives, *P. amazonicus* should be compared to *Ph. caribaeo-quercicola*, its closest kin, and to *Phellinus* sp 1. The main morphological and ecological features of *P. amazonicus* also educe *P. gabonensis*, a species spanning over the western edge of the Guineo-Congolian rainforest.

Phellinus amazonicus and *P. caribaeo-quercicola* differ in slight morphological features, such as the gross habit of the basidiomata. The upper or lateral indurated margins become rimose with age in *P. caribaeo-quercicola*,

while it remains smooth in *P. amazonicus*. More obviously, both species differ in their ecological requirements in terms of substrate, host specificity/preference, habitat, and as a consequence, distribution range. *Phellinus caribaeo-quercicola* is known growing primarily on living trunks or branches of *Quercus* spp. (Fagaceae). It has originally been described from monospecific *Q. cubana* A. Rich (Fagaceae) or mixed *Q. cubana-Pinus* (Pinaceae) stands in western Cuba (Decock et al. 2005). Since then, it has been found on *Quercus* sp. in South Florida, USA (Vlasák et al. 2011). Oak forests from Cuba and South Florida are open habitat with distant trees. Marked rain seasonality with a 6 mo dry period characterized the local climate. Both the habitat and precipitation regime are different from the hyper humid rainforests of French Guiana and Ecuador.

Phellinus amazonicus and *P. gabonensis* are very comparable, both in their macro- and micro-morphology and ecology (type of substrate and habitat). They are hardly distinguishable on the basis of their morphology and are distinguished by their geographical distribution and genetic divergence.

Phellinus amazonicus could be also compared to the East Asian species *P. castanopsidis* and *P. ellipsoideus* (Cui and Dai 2008). *Phellinus ellipsoideus* forms extended, continuous pulvinate sheets underneath large, dead, fallen trunks (Cui and Dai 2008), which feature distinguishes it from the compact, dense, well-delimited cushion-shaped basidiomata of *P. amazonicus*. *Phellinus castanopsidis* forms pulvinate basidiomata on living trunks of *Castanopsis*, a subtropical, Asian Fagaceae (Cui and Decock 2013). Both *P. ellipsoideus* and *P. castanopsidis* are known from subtropical East Asia (Southern China and Northern Thailand).

Phellinus sp. 1.

This species has been observed few times, notwithstanding its remarkable basidiomata, and should be rare. It is known with certainty from a single collection originating from eastern Argentina (FIG. 1). Two specimens originating from the Atlantic forest (subtropical rainforest phytogeographical area) in Southern Brazil (Iganci et al. 2011) might pertain to this taxon, but no DNA sequence is available.

The morphological distinctions between *P. amazonicus* and from *Phellinus* sp. 1 are still uncertain and would need confirmation when more material of the latter is available. Microscopically, we could not find any unequivocal character that could be used to separate *Phellinus* sp. 1 and *P. amazonicus*. The shape and size ranges of all classical microscopic characters overlap between specimens of both clades. Nevertheless, *Phellinus* sp. 1 could differ macroscopically, in developing extended and continuous pulvinate sheets on dead, standing or fallen trunks. A similar development is observed for instance in *P. ellipsoideus* (Cui and Dai 2008). This contrasts with the smaller, well-delimited, cushion-shaped basidiomata of *P. amazonicus*. The distribution range of both taxa might also separate them, but this should still be ascertained. More collections and sets of DNA sequence data are desirable before describing this species.

Specimens examined.—ARGENTINA. MISIONES PROVINCE: Parque Nacional Iguazú, approx 25°41′43″S, 54°26′12″W, on a dead trunk, *Ocotea* sp. (Lauraceae), Aug 2008, *M. Amalfi* (MUCL 51334, culture ex- MUCL 51334). BRASIL. RIO GRANDE DE SUL STATE: Dom Pedro de Alcântara, approx 29°22′10″S, 49°50′59″W, on a dicotyledonous dead wood, unidentified angiosperm, 12 Mar 2010, *Campos Santana 013/10*, (ICN); SANTA CATARINA STATE: Itapuá, Reserva Particular do Patrimônio Natural (RPPN) de Volta Velha, approx 26°04′05″S, 48°37′30″W, on a dicotyledonous dead wood, unidentified angiosperm, 23 Feb 2011, *Campos Santana 515/11* and *516/11* (ICN); ibid., on a dicotyledonous dead wood, unidentified angiosperm, 29 Apr 2013, *Campos Santana 655/13* (ICN).

Phellinus setulosus (Lloyd) Imazeki, Bull. Natl. Sci. Mus. Tokyo 6:104. 1943.
MycoBank MB336263

≡ *Fomes setulosus* Lloyd, Synopsis of the genus *Fomes* 7:243, 1915. MycoBank MB336263

Phellinus setulosus was described in 1915 by Lloyd (as *Fomes setulosus* Lloyd) on the basis of a collection from Sri Lanka. Corner (1991) and Robledo et al. (2003) redescribed this species in many details and highlighted its main morphological features: pileate habit, often with large (up to massive) pileus, dimitic hyphal system with short skeletal hyphae, apically curved to hamate hymenial setae and broadly ellipsoid, whitish to pale yellowish basidiospores. Most of these microscopic features are shared by the species of the *P. caribaeo-quercicola* complex, as highlighted by Decock et al. (2007) and Yombiyeni et al. (2011). Notably, the short skeletal hyphae are present in *P. caribaeo-quercicola*, *P. gabonensis* (Decock et al. 2007, Yombiyeni et al. 2011) and *P. amazonicus* (FIG. 5). Decock et al. (2007) and Yombiyeni et al. (2011) therefore suggested possible affinities of *P. setulosus* with *P. caribaeo-quercicola* and *P. gabonensis*.

Phellinus setulosus is a Southeast Asian species reported southward down to Australia and New Zealand (Cunningham 1965, Corner 1991). As demonstrated above, confusions with *F. wahlbergii* occurred (cf. above). Yet, previously, Buchanan and Ryvarden (2000) and Robledo et al. (2003) confirmed that reports of *P. setulosus* from New Zealand (Cunningham 1963) were based on specimens that in fact, were shown to pertain to *F. wahlbergii*.

The species is also reported from the Neotropics (Ryvarden 204) but the supporting voucher specimens

should be critically re-evaluated. Misidentifications with *P. amazonicus* are not to be excluded.

Diversity within the Neotropical Phellinus *species with pulvinate basidiomata and hooked setae.*—Previous works have shown the occurrence of so-called "cryptic species" in the Hymenochaetales. These species have no or few "indication[s] of perceptible morphological change" (Fischer and Binder 2004). They are best evidenced by molecular data, considered alone (phylogenetic species concept/recognition) or linked to ecological or biogeographical data (e.g. Fischer 2002; Fischer and Binder 2004; Fischer et al. 2005; Decock et al. 2007; Amalfi et al. 2010; Amalfi and Decock 2013; Campos-Santana et al. 2014).

In our case, the basidiomata of *P. amazonicus* is, as far as we could apprehend its morphology, identical to the basidiomata of *P. gabonensis* (Yombiyeni et al. 2011); they are sibling species. This impedes to define unequivocal morphospecies concepts. Furthermore, *P. amazonicus* and *P. gabonensis* occupy analogous ecological niches in their respective rainforest ecosystem (Amazonian and Guineo-Congolian), therefore also impeding the definition of ecological species concepts. They form a morpho-ecological complex with identical morphological traits in analogous environments (see also Yombiyeni et al. 2015). Nevertheless, given their genetic and distribution specificities, a specific taxonomic status is proposed for the collections from French Guiana, Ecuador and Trinidad.

As well, the basidiomata of *P. caribaeo-quercicola* and of *Phellinus* sp. 1 are also, in their micromorphology, identical to those of *P. amazonicus*. These species slightly differ in their macromorphology and more obviously in their autecology, including habitat and geographic distribution. Proposing a specific status for *Phellinus* sp. 1. would be valuable.

Gilbertson and Ryvarden (1987) noted that within the *Fomitiporia robusta* complex, speciation could have resulted from "(physiological) adaptation to different substrates and vastly different environmental factors". The multiplicity of phylogenetic species within the *F. robusta* complex showed using multiple DNA loci and having differential ecological requirements (Amalfi et al. 2012; Vlasák and Kout 2011) supports this hypothesis, as it might for *P. caribaeo-quercicola*, *P. amazonicus* and *Phellinus* sp 1.

Amalfi et al. (2012) indicated the need to consider a combination of multiple complementary descriptors for species description in Hymenochaetaceae. These descriptors should include as a rule, morphological data, genomic (multilocus), ecological and biogeographical data, and in the best case, biological data to define a more "holistic", "Integrated" or "Consolidated" species concept (Puillandre et al. 2012; Quaedvlieg et al. 2014). This should certainly be extended to poroid Hymenochaetaceae. However, still few ecological parameters are systematically collected to complement the species concept.

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LITERATURE CITED

Amalfi M, Decock C. 2013. *Fomitiporia castilloi* sp. nov. and evidence for multiples clades around *F. apiahyna* in Meso- and South America, representing potential species. Mycologia 105:873–887, doi:10.3852/11-423

———, ———. 2014. *Fomitiporia expansa*, an undescribed species from French Guiana. Cryptogam, Mycol 35: 73–85, doi:10.7872/crym.v35.iss1.2014.73

———, Raymundo T, Valenzuela R, Decock C. 2012. *Fomitiporia cupressicola* sp. nov., a parasite on *Cupressus arizonica*, and additional unnamed clades in the southern USA and northern Mexico, determined by multilocus phylogenetic analyses. Mycologia 104:880–893, doi: 10.3852/11-196

———, Robledo G, Decock C. 2014. *Fomitiporia baccharidis* comb. nov., a little known species from high elevation Andean forests and its affinities within the Neotropical *Fomitiporia* lineages. Mycol Prog 13:1075–1087, doi: 10.1007/s11557-014-0995-x

———, Yombiyeni P, Decock C. 2010. *Fomitiporia* in sub-Saharan Africa: morphology and multigene phylogenetic analysis support three new species from the Guineo-Congolian rainforest. Mycologia 102:1303–1317, doi:10.3852/09-083

Buchanan PK, Ryvarden L. 2000. An annotated checklist of polypore-like fungi recorded from New Zealand.

New Zeal J Bot 38:265–323, doi:10.1080/0028825X.2000.9512683

Campos-Santana M, Amalfi M, Robledo G, Borges da Silveira RM, Decock C. 2014. *Fomitiporia neotropica*, a new species from South America evidenced by multilocus phylogenetic analyses. Mycol Prog 13:601–615, doi:10.1007/s11557-013-0943-1

———, Robledo G, Decock C, Borges da Silveira RM. 2015. Diversity of the poroid Hymenochaetaceae (Basidiomycota) from the Atlantic Forest and Pampa in Southern Brazil. Cryptogam Mycol 36:43–78, doi:10.7872/crym.v36.iss1.2015.43

———, Silveira RMB. 2011. *Phellinus gabonensis* (Hymenochaetaceae, Agaricomycota): In: VII Congreso Latinoamericano de Micología, 2011, Costa Rica. Resúmenes y Programa Latino-Micología innovación y progreso VII Congreso Latinoamericano de Micología 1:71.

Castresana J. 2000. Selection of conserved blocks from multiple alignments for their use in phylogenetic analysis. Mol Biol Evol 17:540–552, doi:10.1093/oxfordjournals.molbev.a026334

Corner EJH. 1991. Ad Polyporaceas VII. The xanthochroic polypores. Nova Hedw Beih 101:1–175, pl. 1, 2.

Cui BK, Day YC. 2008. Wood-rotting fungi in eastern China 2. A new species of *Fomitiporia* from Wanmulin Nature Reserve, Fujian Province. Mycotaxon 105:343–351.

———, Decock C. 2013. *Phellinus castanopsidis* sp. nov. (Hymenochaetales) from Southern China, with preliminary phylogeny based on rDNA sequences. Mycol Prog 12:341–351, doi:10.1007/s11557-012-0839-5

Cunningham GH. 1963. The Thelephoraceae of Australia and New Zealand. Bull NZ Dep Sci Ind Res 145:1–359.

———. 1965. Polyporaceae of New Zealand. Bull NZ Dep Sci Ind Res 64:1–304.

Decock C, Amalfi M, Robledo G, Castillo G. 2013. *Phylloporia nouraguensis* (Hymenochaetales, Basidiomycota), an undescribed species from the Neotropics. Cryptogamie Mycol 34:15–27, doi:10.782/crym.v34.iss1.2013.15

———, Bitew A, Castillo G. 2005. *Fomitiporia tenuis* and *Fomitiporia aethiopica* (Basidiomycetes, Hymenochaetales), two undescribed species from the Ethiopian Highlands: taxonomy and phylogeny. Mycologia 97:124–132, doi:10.3852/mycologia.97.1.121

———, Herrera Figueroa S, Robledo G, Castillo G. 2006. *Phellinus caribaeo-quercicola* sp. nov., parasitic on *Quercus cubana*: taxonomy and preliminary phylogenetic relationships. Mycologia 98:265–274, doi:10.3852/mycologia.98.2.265

———, ———, ———, ———. 2007. *Fomitiporia punctata* (Basidiomycota, Hymenochaetales) and its presumed taxonomic synonyms in America: taxonomy and phylogeny of some species from tropical/subtropical areas. Mycologia 99:733–752, doi:10.3852/mycologia.99.5.733

de Granville JJ. 1986. Les formations végétales de la bande côtière de Guyane Française. In: Le littoral côtier guyanais, fragilité de l'environnement. 1er Congrès régional de l'environnement, Sepanguy, Cayenne. p 47–63.

———. 1988. Phytogeographical characteristics of the Guianan forest. Taxon 37:578–594, doi:10.2307/1221101

———. 2002. Milieux et formations végétales de Guyane. Acta Bot Gallica 149:319–337, doi:10.1080/12538078.2002.10515965

Eva HD, Glinni A, Janvier P, Blair-Myers C. 1999. Vegetation map of tropical South America at 1 :5 000 000. Space Application Institute, Global Vegetation Monitoring Unit, TREES Publ. Series D:1–44.

Fischer M. 2002. A new wood-decaying basidiomycete species associated with esca of grapevine: *Fomitiporia mediterranea* (Hymenochaetales). Mycol Prog 1:315–324, doi:10.1007/s11557-006-0029-4

———, Binder M. 2004. Species recognition, geographic distribution and host-pathogen relationships: a case study in a group of lignicolous Basidiomycetes, *Phellinus* s.l. Mycologia 96:799–811, doi:10.2307/3762113

Fischer M, Edwards J, Cunnington JH, Pascoe IG. 2005. Basidiomycetous pathogens on grapevine: a new species from Australia—*Fomitiporia australiensis*. Mycotaxon 92:85–96.

Gilbertson RL, Ryvarden L. 1987. North American Polypores. Part 2. *Megasporoporia–Wrightoporia*. Oslo, Norway: Fungiflora. 449 p.

Iganci JRV, Heiden G, Miotto STS, Pennington RT. 2011. Campos de Cima da Serra: the Brazilian Subtropical Highland Grasslands show an unexpected level of plant endemism. Bot J Linn Soc 167:378–393, doi:10.1111/j.1095-8339.2011.01182.x

Kirk PM, Cannon PF, David JC, Stalpers JA. 2001. Ainsworth & Bisby's Dictionary of the Fungi, 9th ed. Wallingford, UK: CABI Publishing. 655 p.

Kornerup A, Wanscher JH. 1981. Methuen handbook of colour. 3rd ed. London: Methuen. 282 p.

Larsen MJ, Cobb-Poulle LA. 1990. *Phellinus* (Hymenochaetaceae). Synopsis Fungorum 3, Oslo, Norway: Fungiflora. 206 p.

Larsson KH, Parmasto E, Fischer M, Langer E, Nakasone KK, Redhead SA. 2006. Hymenochaetales: a molecular phylogeny for the hymenochaetoid clade. Mycologia 98:926–936, doi:10.3852/mycologia.98.6.926

Lloyd GC. 1915. Synopsis of the genus *Fomes*. Mycol Writ 4:209–288.

Lowe JL.1957. Polyporaceae of North America. The genus *Fomes*. Syracuse, New York: State Univ. College of Forestry, Syracuse Univ. 97 p.

Maddison D, Maddison W. 2000. MacClade 4.0. Analysis of phylogeny and character evolution. Sunderland, Massachusetts: Sinauer Associates.

Matheny PB, Wang Z, Binder M, Curtis JM, Lim YW, Nilsson RH, Hughes KW, Petersen RH, Hofstetter V, Ammirati JF, Schoch C, Langer GE, McLaughlin DJ, Wilson AW, Crane PE, Frøslev T, Ge ZW, Kerrigan RW, Slot JC, Vellinga EC, Liang ZL, Aime MC, Baroni TJ, Fischer M, Hosaka K, Matsuura K, Seidl MT, Vaura J, Hibbett DS. 2007. Contributions of *rpb2* and *tef1* to the phylogeny of mushrooms and allies (Basidiomycota, Fungi). Mol Phylogenet Evol 43:430–451, doi:10.1016/j.ympev.2006.08.024

Murrill WA. 1907. (Agaricales) Polyporaceae. N Am Flora 9:1–72.

Niemelä T. 1975. On Fennoscandian polypores. IV. *Phellinus igniarius*, *P. nigricans* and *P. populicola* n. sp. Ann Bot Fenn 12:93–122.

Peel MC, Finlayson BL, McMahon TA. 2007. Updated world map of the Köppen-Geiger climate classification. Hydrol Earth Syst Sc 11:1633–1644, doi:10.5194

Pieri M, Rivoire B. 2000. Le genre *Phellinus*. Quelques espèces rares ou critiques en France, avec une clé des espèces du genre *Phellinus s.l.* signalées en Europe occidentale. Bull Soc Mycol Fr 116:305–331.

Posada D, Crandall KA. 1998. Modeltest: testing the model of DNA substitution. Bioinformatics 14:817–818, doi:10.1093/bioinformatics/ 14.9.817

Puillandre N, Lambert A, Brouillet S, Achaz G. 2012. ABGD, Automatic Barcode Gap Discovery for primary species delimitation. Mol Ecol 21:1864–1877, doi:10.1111/j.1365-294X.2011.05239.x

Quaedvlieg W, Binder M, Groenewald JZ, Summerell BA, Carnegie AJ, Burgess TI, Crous PW. 2014. Introducing the Consolidated Species Concept to resolve species in the Teratosphaeriaceae. Persoonia 33:1–40, doi:10.3767/003158514X681981

Rambaut A, Drummond AJ. 2007. Tracer 1.4. http://beast.bio.ed.ac.uk/Tracer).

Rehner SA, Buckley E. 2005. A *Beauveria* phylogeny inferred from nuclear ITS and EF1-a sequences: evidence for cryptic diversification and links to *Cordyceps* teleomorphs. Mycologia 97:84–98, doi:10.3852/mycologia.97.1.84

Rizzo DM, Gieser PT, Burdsall HH. 2003. *Phellinus coronadensis*: a new species from southern Arizona, USA. Mycologia 95:74–79, doi:10.2307/ 3761963

Robledo G, Urcelay C, Rajchenberg M. 2003. New species causing decay on living *Polylepis australis* in Cordoba, central Argentina. Mycologia 95:347–353, doi:10.2307/3762046

Ronquist F, Teslenko M, van der Mark P, Ayres D, Darling A, Hōhna S, Larget B, Liu L, Suchard MA, Huelsenbeck JP. 2012. MrBayes 3.2: efficient Bayesian phylogenetic inference and model choice across a large model space. Syst Biol 61:539–542, doi:10.1093/sysbio/sys029

Ryvarden L. 1990. Type studies in the Polyporaceae – 22. Species described by C. G. Lloyd in Polyporus. Mycotaxon 38:83–102.

———. 2004. Neotropical Polypores 1. Introduction, Ganodermataceae and Hymenochaetaceae. Syn Fung 19:1–229.

———, Johansen I. 1980. A preliminary polypore flora of East Africa. Oslo: Fungiflora. 636 p.

Simmons MP, Pickett KM, Miya M. 2004. How meaningful are Bayesian support values? Mol Biol Evol 21:188–199, doi:10.1093/molbev/msh014

Stamatakis A, Hoover P, Rougemont J. 2008. A rapid bootstrap algorithm for the RAxML Web-Servers. Syst Biol 57:758–771, doi:10.1080/10635150802429642

Swofford DL. 2003. PAUP* 4. Phylogenetic analysis using parsimony (*and other methods). Sunderland: Massachusetts. Sinauer Associates.

Taylor JW, Geiser DM, Jacobson DJ, Kroken S, Kasuga T, Geiser DM, Hibbett DS, Fisher MC. 2000. Phylogenetic species recognition and species concepts in fungi. Fungal Genet Biol 31:21–32, doi:10.1006/fgbi.2000.1228

———, Turner E, Pringle A, Dettman J, Johannesson H. 2006. Fungal species: thoughts on their recognition, maintenance, and selection. In: Gadd GM, Watkinson SC, Dyer PS, eds. Fungi in the Environment. p 313–339.

Thiers B. (continuously updated). Index Herbariorum: a global directory of public herbaria and associated staff. New York Botanical Garden's Virtual Herbarium. http://sweetgum.nybg.org/ih/

Thompson JD, Gibson TJ, Plewniak F, Jeanmougin F, Higgins DG. 1997. The CLUSTAL_X windows interface: flexible strategies for multiple sequence alignment aided by quality analysis tools. Nucleic Acids Res 25:4876–4882, doi:10.1093/nar/25.24.4876

Tian XM, Yu HY, Zhou LW, Decock C, Vlasák J, Dai, YC. 2013. Phylogeny and taxonomy of the *Inonotus linteus* complex. Fungal Divers 58:159–169, doi:10.1007/s13225-012-0202-9

Tomšovský M, Sedlák P, Jankovský L. 2010a. Species recognition and phylogenetic relationships of European *Porodaedalea* (Basidiomycota, Hymenochaetales). Mycol Prog 9:225–233, doi:10.1007/s11557-009-0628-y

———, Vampola P, Sedlák P, Byrtusová Z, Jankovský L. 2010b. Delimitation of central and northern European species of the *Phellinus ignarius* group (Basidiomycota, Hymenochaetales) based on analysis of ITS and translation elongation factor 1 alpha DNA sequences. Mycol Prog 9:431–445, doi:10.1007/s11557-009-0653-x

Valencia R, Condit R, Foster RB, Romoleroux K, Villa Muñoz G, Svenning JC, Magård E, Bass M, Losos LC, Balslev H. 2004. Yasuni forest Dynamics plot, Ecuador. In: Losos EC, Leigh J, Giles E, eds. Tropical forest diversity and dynamism: findings from a large-scale plot network. Chicago: Univ. Chicago Press. p 609–628.

Vlasák J, Kout J. 2011. Pileate *Fomitiporia* species in the USA. New combinations *Fomitiporia calkinsii* and *F. bakeri*. Mycol Prog 10:445–452, doi:10.1007/s11557-010-0715-0

———, ———, Vlasák Jr J, Ryvarden L. 2011. New records of polypores from southern Florida. Mycotaxon 118:159–176, doi:10.5248/118.159

———, Li HJ, Zhou LW, Dai YC. 2013. A further study on *Inonotus linteus* complex (Hymenochaetales, Basidiomycota) in tropical America. Phytotaxa 124:25–36, doi:10.11646/phytotaxa.124.1.3

Wagner T, Fischer M. 2002. Proceeding toward a natural classification of the worldwide taxa *Phellinus* s.l and *Inonotus* s.l. and phylogenetic relationship of allied genera. Mycologia 94:998–1016, doi:10.2307/3761866

White TJ, Bruns T, Lee S, Taylor JW. 1990. Amplification and direct sequencing of fungal ribosomal RNA genes for phylogenetics: In: Innis MA, Gelfand DH, Sninsky JJ, White TJ, eds. PCR Protocols: a guide to methods and applications. New York: Academic Press. p 315–322.

Yombiyeni P, Amalfi M, Douanla-Meli C, Decock C. 2011. Poroid Hymenochaetaceae from Guineo-Congolian rainforest: *Phellinus gabonensis* sp. nov. from Gabon—taxonomy and phylogenetic relationships. Mycol Prog 10:351–362, doi:10.1007/s11557-010-0708-z

———, Balezi A, Amalfi M, Decock C. 2015. Hymenochaetaceae from the Guineo-Congolian rainforest: three new

species of *Phylloporia* based on morphological, DNA sequences and ecological data. Mycologia 107:996–1011, doi:10.3852/14-298

Zhou LW, Vlasák J, Decock C, Assefa A, Stenlid J, Abate D, Wu SH, Dai YC. 2015. Global diversity and taxonomy of the *Inonotus linteus* complex (Hymenochaetales, Basidiomycota): *Sanghuangporus* gen. nov., *Tropicoporus excentrodendri* and *T. guanacastensis* gen. et spp. nov., and 17 new combinations. Fungal Divers 77:335–347. doi:10.1007/s13225-015-0335-8

List of specimens examined. ECUADOR. ORELLANA PROVINCE: Yasuni Biosphere reserve / Yasuni National Park, in the vicinity of the Biological Station, approx. 0°41' S – 76°24' W, sendero "Mirador", on a dead fallen trunk, approx. 50 cm diam, unidentified angiosperm, 01 Jul 2008, *C. Decock, EC-08-49* (MUCL 51476; culture ex- MUCL 51476); ibid., on a dead fallen trunk, approx. 80 cm diam, unidentified angiosperm, *C. Decock, EC-08-50* (MUCL51477; culture ex-MUCL51477); ibid., on a dead fallen trunk, unidentified angiosperm, *C. Decock, EC-08-51* (MUCL 51478; culture ex- MUCL 51478); ibid., *C. Decock, EC-08-52* (MUCL; culture ex- MUCL); ibid. on a dead fallen trunk, approx. 80 cm diam, unidentified angiosperm, 03 Jul 2008, *C. Decock, EC-08-62* (MUCL 51483; culture ex- MUCL 51483); ibid., Yasuni CTFS-ForestGEO forest monitoring plot, on a dead fallen trunk, unidentified angiosperm, approx. 80 cm diam, 04 Jul 2008, *C. Decock, EC-08-70* (MUCL 51487; culture ex- MUCL 51487). FRENCH GUIANA. Municipality of Regina, Nouragues Natural Reserve, CNRS "inselberg" research station, approx. 4°05' N – 52°41'W, Petit Plateau, on a dead standing trunk, broken at about 4 m high, unidentified angiosperm, from the base up to approx. 1 m high, 30 Jul 2010, *C. Decock, FG-10-172* (MUCL 53056, culture ex- MUCL 53056); ibid., Petit Plateau, on a dead standing trunk, approx. 50 cm diam, unidentified angiosperm, 01 Aug 2010, *C. Decock, FG-10-217* (MUCL 53084, culture ex- MUCL 53084); ibid., Petit Plateau, fallen trunk, underneath, unidentified angiosperm, 02 Aug 2010, *C. Decock, FG-10-222* (MUCL); ibid., Petit Plateau, on a dead stump, unidentified angiosperm, 02 Aug 2010, *C. Decock, FG-10-234* (MUCL 53095, culture ex- MUCL 53095); ibid., Grand Plateau, dead fallen trunk, approx. 80-90 cm diam, unidentified angiosperm, 04 Aug 2010, *C. Decock, FG-10-269* (MUCL 53117, culture ex- MUCL 53117); ibid., Petit Plateau, on a dead stump, approx. 90 cm diam, 06 Aug 2010, *C. Decock, FG-10-282* (MUCL); ibid., Petit Plateau, on a dead fallen trunk, unidentified angiosperm, approx. 60-70 cm diam, 06 Aug 2010, *C. Decock, FG-10-288* (MUCL 53128, culture ex- MUCL 53128); ibid., on the way to the so-called terrasses, at the Nouragues inselberg, on a dead fallen trunk, 10 Aug 2010, *C. Decock, FG-10-326* (MUCL 53141, culture ex- MUCL 53141); ibid., Petit Plateau, approx. intersection of tracks (layons) 21 & G, at the base of a living trunk, *Minguartia guianensis* (Olacaceae), 28 Jun 2011, *C. Decock, FG-11-378* (MUCL); ibid., on the way to the so-called terrasses, Nouragues inselberg, on a dead piece of wood, approx. 40 cm diam, 29 Jun 2011, *C. Decock, FG-11-*

422 (MUCL 53686; culture ex- MUCL 53686); ibid., Grand Plateau, K-L × 15-16, on a dead fallen branch, approx. 30 cm diam, unidentified angiosperm, 03 Jul 2011, *C. Decock, FG-11-501* (MUCL 53722, culture ex- MUCL 53722); on a dead standing trunk, at the base, 16 Jul 2013, *C. Decock, FG-13-751* (MUCL 55075, culture ex- MUCL 55075); along the track to the Inselberg Savane Roche Virginie, approx. 04.189°N – 052.143° W, elev. approx. 80 masl, on a dead fallen trunk with mosses, 07 Apr 2014, *C. Decock, FG-14-818* (MUCL 55283, culture ex- MUCL 55283); CNRS "Pararé" research station, approx. 4°02’ N – 52°41’W, elev. approx. 130 masl, on a dead fallen trunk, approx. 1.0 m diam, 07 Apr 2015, *C. Decock, FG-15-901* (MUCL 55617, culture ex- MUCL 55617.); ibid., on a dead fallen trunk, approx. 80 cm diam, 09 Apr 2015, *C. Decock, FG-15-934* (MUCL55627, culture ex- MUCL 55627); ibid., on a dead fallen trunk, approx. 80 cm diam, 12 Apr 2015, *C. Decock, FG-15-956* (MUCL55663, culture ex- MUCL 55663); ibid., on a dead fallen trunk, approx. 30 cm diam, 14 Apr 2015, *C. Decock, FG-15-974* (MUCL); Municipality of Maripasoula, Marouini river, approx. 02.731°N – 054.0161° W, elev. approx. 135 masl, forest, on a dead fallen trunk, approx. 50-60 cm diam, 11 Jul 2013, *C. Decock, FG-13-720* (MUCL 55050, culture ex- MUCL 55050); ibid, dead fallen trunk, underneath, *C. Decock, FG-13-729* (MUCL 55053, culture ex- MUCL 55053); Municipality of Awala Yalimapo, Réserve Naturelle Amana, on a dead fallen trunk, unidentified angiosperm, 15 Apr 2014, *C. Decock, FG-14-860* (MUCL 55321, culture ex- MUCL 55321); approx. 05°4699’ N – 053°5728’W, elev. approx. 60 masl, dead fallen trunk and roots, possibly *Dimorphandra polyandra* (Caesalpiniaceae), 17 Apr 2014, *C. Decock, FG-14-893* (MUCL 55326, culture ex- MUCL 55326). TRINIDAD. Brazil, forest, 06 Mar 1921, F.J. Seaver, 3065 (NYBG); Mora forest, east of Sangre Grande, 10 Apr 1921, *F.J. Seaver*, without number (NYBG).